PROREADY
Ready To Use Culture Media

CE IVD

UPUTSTVO ZA UPOTREBU (SRB)

Mueller Hinton Agar Plate No. 2

Podloga za ispitivanje osetljivosti mikroorganizama na antimikrobne agense sa niskim sadržajem timina, timidina, kalcijuma i magnezijuma.

Sadržaj pakovanja:

Šifra artikla (pakovanja) REF	Opis	Šifra primarnog pakovanja:	Broj podloga
PRM1084V20	Podloga izlivena u petri posudama od ø90	**PRM1084**	**20**
PRM1084V60			**60**
PRM1084V240			**240**
PRM1084M40	Podloga izlivena u petri posudama od ø50		**40**

Uputstva

Pod aseptičnim uslovima se standardna suspenzija test organizma nanosi (obično brisom) preko cele površine podloge.

Princip i interpretacija

Cilj testiranja antimikrobne osetljivosti je da se in vitro procenom predvidi verovatnoća uspešnog lečenja pacijenta od infekcija određenim antimikrobnim agensom (1). Formulacija Mueller Hintona prvobitno je razvijena kao jednostavna, prozirna podloga za kultivaciju patogenih Neisseria spp. (2). Zatim su razvijene druge podloge koje su zamenile upotrebu Mueller Hinton Agara za kultivaciju patogenih Neisseria spp., ali je ona postala široko korišćena u određivanju rezistencije gonokoka i drugih organizama na sulfonamide. Mueller Hinton Agar se sada koristi kao podloga za testiranje antimikrobne osetljivosti (3). Mueller Hinton Agar se preporučuje za izvođenje disk-difuzione metode koja podrazumeva difuziju antimikrobnog agensa impregniranog na papirnom disku kroz agarizovanu podlogu, kao što je opisano u CLSI odobrenom standardu (4). Mueller Hinton Agar je izabran od strane CLSI zbog nekoliko razloga:

I. Pokazuje dobru reprodukciju od serije do serije u testovima antimikrobne osetljivosti.
II. Ima niske vrednosti inhibitora sulfonamida, trimetoprima i tetraciklina.
III. Podržava rast većine nezahtevnih bakterijskih patogena.
IV. Evidentirano je mnogo podataka i iskustva o njegovom učinku (9).

Mueller Hinton Agar No. 2 se koristi u testiranju osetljivosti brzo-rastućih aerobnih i fakultativno anaerobnih bakterija iz kliničkih uzoraka. Kirby-Bauer i ostali su preporučili ovu podlogu za izvođenje testova osetljivosti na antibiotike korišćenjem jednog diska visoke koncentracije (5). WHO komitet za standardizaciju je prihvatilo Mueller Hinton Agar kao podlogu za određivanje antimikrobne osetljivosti organizama zbog njegove reproduktivnosti (6). Podloga je dizajnirana sa niskim sadržajem timina i timidina i koncentracijama jona kalcijuma i magnezijuma koje su preporučene od strane CLSI (3). Timin i timidin ihibiraju sulfonamide i trimethoprim (9,10), a kalcijum i magnezijum (11,12) utiču na aktivnost aminoglikozidnih antibiotika. Podloga nije preporučena za zahtevne organizme. Goveđi infuzum i kiselinski hidrolizat kazeina obezbeđuje azotna jedinjenja, ugljenik, sumpor i druge esencijalne hranljive materije. Skrob deluje kao zaštitni koloid protiv toksičnih supstanci prisutnih u podlozi. Hidrolizom skroba dobija se dekstroza koja sluzi kao izvor energije. Ovi sastojci su izabrani zbog niskog sadržaja timina i timidina što je značajno prilikom određivanja MIC vrednosti za Enterococcus fecalis sa sulfametaksazoltrimetoprimom (SXT). Koncentracije jona kalcijuma i magnezijuma su prilagođene kako bi se obezbedile količine koje preporučuje CLSI, važne za dobijanje tačne MIC vrednosti u slučaju aminoglikozida i Pseudomonas aeruginosa (3). Postupak Kirby-Bauer-a je zasnovan na difuziji antimikrobnih agenasa impregniranih na papirnim diskovima u agarizovanu podlogu. Ovaj metod koristi disk sa jednom koncentracijom antimikrobnog agensa, a diametri zona inhibicije su u korelaciji sa vrednostima minimalne inhibitorne koncentracije (MIC) (2,3,7). Standardna suspenzija mikroorganizama se brisom nanosi preko cele površine podloge. Papirni diskovi impregnirani određenim količinama antimikrobnih agenasa se onda stavljaju na površinu podloge, inkubiraju i zone inhibicije se mere oko svakog diska. Osetljivost se određuje upoređivanjem sa CLSI standradima (8). Faktori koji utiču na ispitivanje osetljivosti disk-difuzionom metodom su: debljina agara, potencijal diska, koncentracija inokuluma, pH podloge i sposobnosti test organizma da produkuje beta-laktamazu (1,8).

Kontrola kvaliteta

Podaci i rezultati kontrole kvaliteta dati su u sertifikatu analize za svaku seriju.

Skladištenje i rok upotrebe

Čuvati između 15-25°C. Nakon prvog otvaranja čuvati na 2-8°C. Upotrebiti pre isteka datuma označenog na nalepnici.

Mere predostrožnosti

Ovaj proizvod ne sadrži hazardne supstance u koncentracijama koje su iznad propisanih limita određenih važećim zakonskim regulativama i zato nije klasifikovan kao opasan. Ipak, preporučeno je slediti smernice iz bezbednosnog lista za pravilnu upotrebu. Ovaj proizvod je namenjen isključivo za upotrebu u laboratorijskim uslovima, od strane profesionalno obučene osobe.
Proizvod ne upotrebljavati ukoliko je primarno pakovanje oštećeno ili proizvod ne odgovara navedenim karakteristikama.

Odlaganje otpada

Odlaganje otpada mora biti u skladu sa nacionalnim i lokalnim regulativama koje su na snazi. Svaka laboratorija je odgovorna za rukovanje i odlaganje otpada koji nastaje u toku rada.

Upotrebljeni simboli

CE	Evropski znak usaglašenosti		Držati uspravno
IVD	In vitro dijagnostičko medicinsko sredstvo	REF	Kataloški broj
	Ne izlagati direktno sunčevim zracima	LOT	Lot broj
	Konsultovati uputstvo za upotrebu		Rok upotebe
	Ne koristiti više puta		Temperatura čuvanja
Σ	Veličina pakovanja		Proizvođač
EC REP	Ovlašćeni predstavnik u Evropskoj uniji		

EC REP	Salus Cons kft. 6722 Szeged, Bécsi krt 23, HUNGARY e-mail: office@saluscons.com

Literatura

1.Murray P. R., Baron J. H., Pfaller M. A., Jorgensen J. H. and Yolken R. H., (Ed.), 2003, Manual of Clinical Microbiology, 8th Ed., American Society for Microbiology, Washington, D.C.
2. Mueller J. H. and Hinton J., 1941, Proc. Soc. Exp. Biol. Med.,48:330.
3. National Committee for Clinical Laboratory Standards, 2000, Approved Standard: M7-A5. Methods for Dilution Antimicrobial Susceptibility Tests for Bacteria that grow aerobically, 5th Ed., NCCLS,Wayne, Pa.
4. NCCLS Approved Standard: ASM-2, 1979, Performance Standards for Antimicrobic disc Susceptibility Tests, 2nd Ed.,
National Committee for Clin. Lab. Standards.
5. Bauer A. W., Kirby W. M., Sherris J. L. and Turck M., 1966, Am. J. Clin. Pathol., 45:493.
6. Present Status and Future Work, WHO Sponsored collaborative study, Chicago, Oct. 1967.
7. Ericsson H. M. and Sherris J. L., 1971, Acta Pathol. Microbiol., Scand. Sect B Suppl., 217:1.
8. National Committee for Clinical Laboratory Standards, 1986, Proposed Standards, M6-P, NCCLS, Villanova, Pa.
9. Koch A. E. and Burchall J. J., 1971, Appl. Microbiol., 22: 812.
10. Ferone R. Bushby R. M., Burchall J. J., Moore W. D., Smith D., 1975, Antimicrob. Agents chemotherap., 7 : 91.
11. Pollock H. M., Minshew B. H., Kenney M. A., Schoenknecht F. D., 1978 , Antimicrob. Agents Chemotherap.; 14:360.
12. DAmato R. F., and Thornsberry C., 1979, Curr. Microbiol., 2 : 135.

Broj rešenja o registraciji: 515-02-02534-22-003

PROREADY
Ready To Use Culture Media

CE IVD

INSTRUCTION FOR USE (EN)

Mueller Hinton Agar Plate No. 2

Medium is used for testing susceptibility of bacteria using antimicrobial agents contain low levels of thymine, thymidine, calcium and magnesium.

Package contents:

Item code (packaging) REF	Description	Primary packaging code:	Number of products
PRM1084V20	Substrate poured into petri dishes of ø90	**PRM1084**	**20**
PRM1084V60			**60**
PRM1084V240			**240**
PRM1084M40	Substrate poured into petri dishes of ø50		**40**

Directions

Standard suspension of test microorganisms aseptically swabbed over the entire surface of the medium.

Principle and interpretation

The goal of susceptibility test is to predict through an *in vitro* assessment the likelihood of successfully treating a patient's infection with a particular antimicrobial agent (1). The Mueller Hinton formulation was originally developed as a simple, transparent agar medium for the cultivation of pathogenic *Neisseria* species (2). Other media were subsequently developed that replaced the use of Mueller Hinton Agar for the cultivation of pathogenic *Neisseria* species, but it became widely used in the determination of sulfonamide resistance of gonococci and other organisms. Mueller Hinton Agar is now used as a test medium for antimicrobial susceptibility testing (3). Mueller Hinton Agar is recommended for the diffusion of antimicrobial agents impregnated on paper disc through an agar gel as described in CLSI (Clinical and Laboratory Standards Institute) Approved Standard (4). Mueller Hinton Agar has been selected by the CLSI for several reasons:

I. It demonstrates good batch-to-batch reproducibility for susceptible testing.
II. It is low in sulfonamide, trimethoprim and tetracycline inhibitors.
III. It supports the growth of most non-fastidious bacterial pathogens.
IV. Many data and much experience regarding its performance have been recorded (9).

Mueller Hinton Agar is used in the susceptibility testing of rapidly growing aerobic and facultatively anaerobic bacteria from clinical specimens. Kirby-Bauer et al recommended this medium for performing antibiotic susceptibility tests using a single disc of high concentration (5). WHO Committee on Standardization of Susceptibility Testing has accepted Mueller Hinton Agar for determining the susceptibility of microorganisms because of its reproducibility (6). The medium is designed to give a low thymine and thymidine content and also the calcium and magnesium ion concentration is adjusted as recommended by CLSI (3). Thymine and thymidine inhibit sulfonamide and trimethoprim (9, 10) activity and calcium and magnesium (11, 12) interferes with the activity of aminoglycoside antibiotics. However, Mueller Hinton Agar No. 2 is not recommended for fastidious organisms. Beef heart infusion and casein acid hydrolysate provide nitrogenous compounds, carbon, sulphur and other essential nutrients. Starch acts as a protective colloid against toxic substances present in the medium. Starch hydrolysis yields dextrose, which serves as a source of energy. These ingredients are selected for low thymine and thymidine content as determined by MIC values for *Enterococcus faecalis* with sulfamethoxazole trimethoprim (SXT). Calcium and magnesium ion concentrations are adjusted to provide the amounts recommended by CLSI to give the correct MIC values with aminoglycosides and *Pseudomonas aeruginosa* (3). The Kirby-Bauer procedure is based on agar diffusion of antimicrobial substances impregnated on paper discs. This method employs disc with a single concentration of antimicrobial agent and the zone diameters observed are correlated with minimum inhibitory concentration (MIC) values (2, 3, 7). A standardized suspension of the organism is swabbed over the entire surface of the medium. Paper discs impregnated with specific amounts of antimicrobial agents are then placed on the surface of the medium, incubated and zones of inhibition around each disc are measured. The susceptibility is determined by comparing with CLSI standards (8). The various factors, which influence disc diffusion susceptibility tests, are agar depth, disc potency, inoculum concentration, pH of the medium and beta-lactamase production by test organisms (1, 8).

Quality control

The data and results of quality control are given in the certificate of analysis for each lot.

Storage and shelf life

Storage between 15-25°C. After opening storage between 2-8°C. Use before expiry date on the label.

Warning and precautions

In vitro diagnostic use only. Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/ face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling clinical specimens. Safety guidelines may be referred in individual safety data sheets.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with clinical sample must be decontaminated and disposed of in accordance with current laboratory techniques.

Symbols used on labels

CE	European Conformity mark		This side up
IVD	is an in vitro diagnostic medical device (IVD)	REF	Catalogue number
	Do not expose directly to sunlight	LOT	Batch code
	Consult instructions for use		Use-by date
	Do not re-use		Temperature limit
	Pack size		Manufacturer
EC REP	European Authorized Representative (Authorised Representative)		

EC REP	Salus Cons kft. 6722 Szeged, Bécsi krt 23, HUNGARY e-mail: office@saluscons.com

Reference

1. Murray P. R., Baron J. H., Pfaller M. A., Jorgensen J. H. and Yolken R. H., (Ed.), 2003, Manual of Clinical Microbiology, 8th Ed., American Society for Microbiology, Washington, D.C.
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5. Bauer A. W., Kirby W. M., Sherris J. L. and Turck M., 1966, Am. J. Clin. Pathol., 45:493.
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